ENTOMOLOGY AND NEMATOLOGY

Panel Manager - Dr. Bruce Campbell, USDA-ARS Albany CA
Program Director - Dr. Mary Purcell-Miramontes

Research grants in this area support studies on plant stress arising from interactions with insects, mites, and nematodes. The goal of the program is to support research which will lead to ways of reducing losses in plant productivity from damage caused by these organisms, and to do so in such a way as to sustain agricultural systems.

Research on plant/insect and plant/nematode interactions is encouraged as is research into the basic biology of these particular invertebrates. Specific emphasis is placed on but not limited to the following areas: (1) mechanisms of plant defense and resistance against insect or nematode attack (biochemical or genetic), (2) insect or nematode growth, development and reproduction (physiological, biochemical or molecular studies), (3) physiological and ecological interactions of biological control agents with their hosts (includes beneficial insects, viruses, bacteria, fungi and other organisms), (4) chemical ecology and behavior, (5) population dynamics (includes evolutionary studies), and (6) fundamental basis of pesticide toxicity and resistance.

2000-01137 Structure and Behavior of Meiotic Holocentric Chromosomes in Hemiptera

Hilburn, L.R.
Arkansas State University; Department of Biological Sciences; State University, AR 72467
Equipment Grant 2000-35302-10156; $24,780; 1 Year

This Equipment Grant is to purchase a research microscope to study meiosis in the Tarnished Plant Bug, *Lygus lineolaris.* Tarnished Plant Bug has holocentric chromosomes; the microtubules that separate them during meiosis attach at numerous points along their length, rather than at one point as is observed in most organisms. Since Tarnished Plant Bug is a pest that damages a wide variety of crops, it is important that its unique form of meiosis be better understood. This microscope will facilitate investigations into how the structure of holocentric chromosomes affects 1) recombination between chromosomes carrying the same genes (homologues) during early meiosis, 2) separation of the homologues in middle meiosis, 3) attachment of microtubules to the chromosomes to insure that the recombined segments are moved to the same end of the cell as the rest of the chromosomes to which they are attached after recombination. The points of exchange of DNA between homologues can be determined by staining the chromosomes with silver and examining them during an early period of meiosis when the homologues are tightly associated with each other. The microtubule attachment points can be seen by treating the chromosomes with fluorescent antibodies that specifically bind either to proteins found in the microtubule attachment points on the chromosome or to proteins of the microtubules. How holocentric structure affects recombination and movement of chromosomes during meiosis are questions whose answers can suggest how fast and how much Tarnished Plant Bug genetics can change to meet new challenges, such as new pesticides, in their environment.

2000-02949 Viral Protein Interactions that Determine the Transmission of Nepoviruses by Nematodes

Gergerich, R.C.; Kim, K.S.; Robbins, R.T.
University of Arkansas, Fayetteville; Department of Plant Pathology, Fayetteville, AR 72701
Strengthening Grant 2001-35302-09980; $150,000; 2 Years

Members within the nepovirus group of plant viruses are transmitted specifically by longidorid nematodes and cause economically important diseases in many crops, especially woody crop plants, many of which are difficult to control. Most plant viruses are spread in nature by a vector, and vector nematodes play an important role in the infection cycle of nepoviruses. An appealing approach for management of plant virus diseases caused by nematode transmitted viruses is to interfere with the interaction between the nematode and the virus that results in virus transmission. In this research we propose to characterize the interaction of nepoviruses with their nematode vectors and to identify the viral and nematode components that are involved in the transmission process. It is our hope that these studies will lay the foundation for development of novel virus disease control methods which target the transmission process. Specifically the objectives for this project are to (1) determine the need for a putative viral helper component in the transmission of tobacco ringspot virus and tomato ringspot virus by *Xiphinema americanum*, (2) determine the role of the viral movement protein, if any, in nematode transmission of tobacco ringspot virus and tomato ringspot virus; (3) identify and isolate the specific nematode factors involved in attachment of virus particles or viral helper component. Knowledge of the biochemical basis for the specific attachment and dissociation of nepoviruses during the transmission process will provide information with the ultimate goal of developing more effective control measures for nepoviruses.

2000-02890 Large Scale Test of Refuges for Managing Pink Bollworm Resistance to Bt Cotton

Carriere, Y.; Dennehy T. J.; Antilla L.; Tabashnik, B.E.
University of Arizona; Department of Entomology; Tucson, AZ 85721-0036
Grant 2001-35302-09976; $220,000; 3 Years

Transgenic crops expressing insecticidal proteins from the bacterium *Bacillus thuringiensis* boost yields and drastically reduce use of synthetic insecticides in some regions of the U.S. Evolution of insect resistance to these crops threatens those benefits. This is especially relevant in Arizona where laboratory selection has easily produced pink bollworm (PBW) resistant to Bt-cotton and selection for resistance is strong with more than 75 % of Bt cotton grown in many regions. The EPA has mandated the refuge/high-dose strategy to delay the evolution of PBW resistance, but information is lacking to develop that strategy. Data on PBW dispersal is needed, and no large-scale field tests of the strategy have been attempted. To address these needs, we will: 1) Map the distribution of Bt and non-Bt cotton fields in Arizona in three successive years, 2) measure dispersal of adult PBW, and 3) determine the relationship between distribution of Bt and non-Bt cotton and frequency of PBW resistance. To achieve these objectives, we will use geographic information system technology (GIS) to integrate statewide data on PBW dispersal, use of Bt and non-Bt cotton, and frequency of PBW resistance. This approach could lead for the first time to empirical determination of guidelines for deployment of transgenic crops, which would have tremendous positive impacts on U.S. agriculture.

2000-02919 Ovarian Dynamics and Host Use in a Tephritid Fly

Papaj, D.R.
University of Arizona; Department of Ecology & Evolutionary Biology; Tucson, AZ 85721
Grant 2001-35302-09977; $213,000; 3 Years

How damaging a pest insect can be to agricultural crops often depends not only on the number of insects attacking the crop but also on the numbers of eggs that females can produce and lay. Knowledge of ovarian development can therefore help us to better forecast a pest's potential for crop damage. One factor that affects how many eggs develop and how fast is the host plant itself. We propose here to examine how egg production by a fruit-feeding insect is influenced by variation in the quality and abundance of the fruit on which its larvae feed. Using the walnut-infesting tephritid fly, *Rhagoletis juglandis,* as a model system, our first objective is to examine how the rate of fruit encounter as well as the quality of fruit encountered influences egg maturation. Our second objective is to examine what fruit stimuli affect ovarian maturation. Our final objective is to use the information obtained on ovarian dynamics to construct an individual-based computer simulation model that predicts levels of infestation over varying conditions of fruit quality and abundance under field or field-cage conditions. This research stands to improve our ability to predict a pest insect population's attack potential over time and space. Information on egg production is pertinent to construction of population models that aid in the development of management strategies such as pesticide spraying and trap deployment schedules. It has implications for sterile release programs, as well as the design and implementation of plant- or fruit-mimicking traps for monitoring or suppressing populations.

2000-02872 Secondary Endosymbionts of Aphids

Purcell, A.H.
University of California, Berkeley; Department Environment Science, Policy & Management-Insect Biology; Berkeley, CA 94720-3112

Grant 2000-35302-9400; $117,000; 2 Years

Aphids are major pests of crops throughout the world and their rapid reproduction is a serious barrier to effective pest management. Certain bacteria, or "primary" symbionts, are necessary for the proper development and reproduction of all aphids. Other bacteria, or "secondary" symbionts, occur less widely in aphids, but also potentially affect reproduction. Our studies of two secondary symbionts of pea aphids, designated PASS and PAR, have shown that they may be harmful or beneficial, depending on environmental conditions. We are particularly interested in the effects of temperature on the success of pea aphids housing different combinations of three bacteria: the primary symbiont, PASS, and PAR. We have experimental evidence that PASS, at least, is able to "rescue" the primary symbiont under temperature conditions of heat stress (short periods of high temperatures) that otherwise completely disrupt the primary symbiosis and aphid reproduction; aphids lacking PASS do not reproduce under these conditions. Our objectives are (1) to definitively localize PASS and PAR in aphids by microscopy and molecular methods in order to better understand the nature of these symbioses, (2) to evaluate the influence of PASS and PAR on aphid reproduction in the absence or incapacitation of the primary symbiont, and (3) to quantify the effects of PASS and PAR on the primary symbiont and on aphid fitness under different temperature regimes. This information is fundamental to understanding how secondary symbionts impact aphid population growth, and may reveal new possibilities for using molecular approaches with bacterial associates of aphids for pest and disease management.

2000-02927 Role of α/β-Hydrolase Fold Enzymes in the Regulation of Juvenile Hormone

Hammock, B.D.
University of California, Davis; Department of Entomology; Davis, CA 95616

Grant 2001-35302-09919; $220,000; 3 Years

Caterpillars or 'worms' are a major target of insecticides. Many existing insect control agents disrupt a regulatory system thus amplifying the effect of the agent in controlling insect pests. Many biological control agents also act by disrupting a regulatory system. This research focuses on one of these regulatory systems that controls the conversion of caterpillars into butterflies or moths. Juvenile hormone (JH) keeps the insect in a feeding stage while loss of JH causes the insect to stop feeding and undergo metamorphosis. This process is partially controlled by two enzymes which degrade JH. In this project we will study the biochemistry of these enzymes and how they regulate metamorphosis. Many techniques will be used for this work including the generation of a picture of the enzyme targets at molecular resolution. We will then apply our basic knowledge to design agents to control key pest insects. We will take two approaches. The more classical approach involves using these degrading enzymes as target for the synthesis of novel insect growth regulators. Because these enzymes are unique to insects we anticipate that the resulting regulators will be very specific for insect pests. The second approach is molecular where we will use biological control agents such as insect viruses to cause the enzymes to be either overproduced or not produced at all. In this way we will generate basic knowledge on insect development that can be utilized by many researchers, and we will apply this information to two different approaches to insect control.

2000-02993 Characterization of Root-knot Nematodes that Infect Resistant Tomato

Williamson, V.M.
University of California, Davis; Department of Nematology; Davis, CA 95616

Grant 2001-35302-10135; $174,420; 3 Years

Root-knot nematodes cause severe damage to many important crops. Although this nematode can infect a broad range of crops, there are considerable differences in the ability of different nematode isolates to reproduce on particular crop cultivars. The goal of this proposal is to identify genetic factors in the nematode that account for this variability. Specifically, a strain of root knot nematodes, VW5, that reproduces on tomato with the nematode resistance gene Mi will be compared to the strain VW4, which can not reproduce on tomato with Mi. Genomic DNA from these two nematode strains will be compared using powerful new selective techniques that allow identification of differences between nearly identical organisms. Once genetic differences are identified, the corresponding genes will be examined and tested to determine whether they are responsible for the ability of strain VW5 to reproduce on tomato with the Mi gene. How these genetic changes allow the nematode to reproduce on resistant tomato will be examined as will the mechanism by which the nematode has acquired the new ability. The proposed research should lead to a better understanding of how root-knot nematodes change their genomes to infect new host plants and should provide insight into strategies for more effective use of natural plant resistance genes for nematode control.

2000-02943 Crystallization of Truncated Bt Cryl Proteins for Improved Efficacy

Federici, B.A.
University of California, Riverside; Department of Entomology; Riverside, CA 92521
Grant 2001-35302-09974; $221,000; 3 Years

Bacterial insecticides based on *Bacillus thuringiensis* (Bt) have provided effective and environmentally safe alternatives to chemical insecticides for control of major lepidopteran pests for more than forty years. However, new chemicals and Bt transgenic plants seriously threaten this valuable technology. If Bt products are to continue to provide alternatives to chemical insecticides, their cost-effectiveness must be improved. In this project, we will use a novel molecular strategy to attempt to improve Bt efficacy by increasing the toxicity per unit weight of Cryl proteins. Cry proteins are the principal active ingredients of Bt insecticides. These proteins can be divided into two halves, a toxic half and a non-toxic half. When only the non-toxic half is produced using genetic engineering technology, it does not crystallize, a property essential for formulation of bacterial insecticides. There are however a few naturally occurring Bt Cry proteins that crystallize well, yet are composed of only the toxic half of the molecule. Thus, in this project we will identify the portions of these smaller proteins that make them crystallize, and then substitute these regions into the toxic portion of truncated Cryl proteins. In addition, we will add genetic enhancer and stabilizing elements to the genetically engineered constructs of truncated Cry proteins to increase their yield and stability. The successful completion of this research has the potential for improving the efficacy of Cryl proteins by approximately 100%. This research will also provide new knowledge about the molecular biology of protein crystallization.

2000-02876 Characterization of Mutants in the Mi-1-Mediated Resistance Pathway

Kaloshian, I.
University of California, Riverside; Department of Nematology; Riverside, CA 92521
Grant 2001-35302-09973; $200,000; 3 Years

Root-knot nematodes are obligate parasites of a wide range of cultivated crops and cause severe yield losses. In tomato, resistance to three major root-knot species in depends on the expression of the Mi-1 gene. Recently, the Mi-1 gene has been isolated by positional cloning and shown to encode a protein product similar to a number of previously cloned plant resistance gene products. However, Mi-1 is a unique gene in that, besides being a nematode resistance gene, it also confers resistance to the potato aphid. We are using a genetic approach to identify the signal transduction components required for the function of Mi-1. This approach involves the generation and identification of mutants with increased disease sensitivity in Mi-1 containing tomato lines. By identifying the ways in which the resistance response becomes defective, will allow us to understand what makes plants resistant. We have currently isolated tomato mutants, generated from Mi-1-containing seeds, with altered nematode resistance. One mutant, *rme-1*, is susceptible to nematodes in spite of the presence of a functional Mi-1 gene. In this project, we intend to characterize and map this mutation. Besides, it is not clear whether the nematode and aphid resistance pathways are identical. We will test whether *rme-1* is also susceptible to the potato aphid. A better understanding of the mechanisms by which resistance genes mediate resistance and identifying components of these pathways, will lead to the ability to engineer durable resistance to these agronomically important pathogens.

2000-03011 Genetic and Biochemical Determinants of Intraspecific Aggression in Argentine Ants
Suarez, A.V.
University of California, San Diego; Department of Biology; La Jolla, CA 92093
Post-doctoral Fellowship; Grant 2000-35302-9417; $90,000; 2 Years

Despite the severe ecological and economic problems associated with the invasions of non-native species, the mechanisms behind their success are often unknown. The invasive Argentine ant, *Linepithema humile*, is a severe agricultural pest in the United States. Within introduced populations, Argentine ants typically maintain expansive super-colonies in which nestmate recognition appears absent. By eliminating costs associated with territoriality, nests of Argentine ants can locate resources more quickly, recruit more workers to resources, and subsequently grow more quickly than the native species they compete with. The purpose of this proposal is to determine the mechanisms behind nestmate recognition in Argentine ants and to examine possible strategies for their control that exploit the low levels of nestmate recognition typical of introduced populations. Specifically, I propose four experiments to examine the relationship between genetic similarity, nestmate recognition, and the chemical cues used in nestmate discrimination in ants. The results of this proposal will help develop control strategies for the control of this widespread and damaging agricultural pest.

2000-02924 Invasion of the Argentine ant: Predicting Rate of Spread
Gordon, D.
Stanford University; Department of Biological Sciences, Stanford, CA 94305-5020
Grant 2001-35302-09981; $170,000; 2 Years

The Argentine ant *Linepithema humile (=Iridomyrmex humilis)* is an invading species that has spread worldwide, and is now a serious pest of citrus in California through its association with homopterans. The Argentine ant has begun to invade the Jasper Ridge Biological Preserve owned by Stanford University, providing a unique opportunity to monitor an invasion before and after it occurs. The study will continue to track the Argentine ants as they expand their range within the reserve. This is the first study to monitor in detail, over a large area, the ecological effects of an invasion of this species. It is not known exactly how Argentine ants invade areas inhabited by native ant species. The study will analyze genetic variation in microsatellite markers to determine whether the ants spread at the invasion front by budding from nearby nests at the front, or instead move in from more distant, inhabited areas where Argentine ants are already established. This work will contribute to future biological control efforts by identifying when rapid expansion of the Argentine ant range is likely, and what conditions slow the spread of this species.

2000-02992 Effective Russian Wheat Aphid Management by Modeling Climate and Natural Enemies
Lester, P.J.; Cipra, J.E.; Peairs, F.B.; Holtzer, T.O.
Colorado State University; Bioagricultural Sciences and Pest Management; Fort Collins, CO 80523-1177
Grant 2001-35302-09896; $160,000; 2 Years

The Russian wheat aphid (RWA) has caused several hundred million dollars damage since its introduction into the US. However, substantial differences in damage occur between years. In Colorado,

$14.3 million damage was estimated for 1997 but only $0.3 million for 1996. Climactic factors, such as snow cover and hours below freezing, play significant roles in this variability. Factors that alter host plant characteristics and the density of natural enemies also influence aphid populations. Our aim is to create a spatial and temporal Geographic Information Systems (GIS) model of RWA and its natural enemies. This model will be used to assess the RWA infestation risk. Risk assessments will be used to tailor management actions. In high-risk areas, the only current effective management tool is resistant wheat. In medium and low risk areas, biological or chemical controls may be acceptable alternatives. Areas where these tools are used instead of resistant wheat will serve as refuges where aphids are not subject to selection pressure associated with resistant wheat. Employing biological and chemical control may reduce genetic changes in aphid populations and prolong the effectiveness of aphid resistant wheat. To gather data for model development and testing, three 5 x 5km sites will be located in areas representing a range of growing conditions. 100 stations will be randomly located within each site. At each station, we will examine the dynamics of RWA and important natural enemies throughout the growing season and winter. GIS layers, including snow cover, temperature, topography and vegetation will be collected.

2000-02995 Male-Biased Sex Ratios and Sex Determination Mechanisms in Parasitoids

Hopper, K.R.; Heimpel, G.E.; Ode, P. J.; Fuester, R.W.
USDA Agricultural Research Service; Beneficial Introductions Research Unit; Newark, DE 19713
Grant 2001-35302-09975; $155,000; 2 Years

Parasitic wasps in the family Braconidae frequently exhibit male-biased sex ratios when mass-reared. Many braconid wasps are important natural enemies of pest insects and these male-biased can reduce the efficiency with which these wasps can be reared and their efficacy once released to control pests. The reason for the male biases is unknown for most species of Braconidae, but the most likely explanation involves a particular mechanism of sex determination. In most wasps, females carry two sets of chromosomes (i.e., are diploid) and males carry only a single set (i.e., are haploid). In some braconids, however, diploid males are produced when individuals carry two copies of one particular gene. Diploid males are inviable or sterile and are primarily produced as a consequence of inbreeding. In this proposal, we test the hypothesis that male-biased sex ratios in the Braconidae can be explained by the production of diploid males. We will establish inbred and outcrossed matings and compare sex ratios and mortality rates resulting from these crosses. In cases where the production of diploid males is suggested, we will confirm their presence using molecular and other genetic tests. If viable adult diploid males are produced, we will determine whether they are capable of mating and whether females mated to diploid males will re-mate with haploid males. These analyses will be carried out for a minimum of 4 species of wasps, and the results will be used to address hypotheses about the evolution of sex determination in braconid wasps.

2000-02954 The Insect Integument: Nature and Nurture: A Tribute to Michael Locke

Willis, J.H.; Riddiford, L.M.
University of Georgia; Department of Cellular Biology; Athens, GA 30602
Conference Grant 2001-35302-09903; $6,600; 1 Year

These funds will be used to support a symposium on the insect integument designed to honor Professor Michael Locke's diverse contributions to fundamental aspects of insect development and physiology. The Symposium will be held in early December 2000 in Montreal, Canada as part of a joint meeting of the Entomological Society of America, The Entomological Society of Canada and Societe d'Entomologie du Quebec. A cadre of international scientists will discuss topics focused primarily on the insect integument including pattern formation, hormonal regulation, assembly and resorption of cuticular components, and the tracheal system. A special feature will be a poster session to be held immediately after the symposium to assure that students, postdoctoral associates and senior participants have the opportunity to interact constructively.

2000-02930 On-site Mixture Processing by Antennal Neurons in Heliothine Moths
Baker, T.C.
Iowa State University; Department of Entomology; Ames, IA 50011
Grant 2001-35302-09886; $156,000; 3 Years

A largely overlooked aspect of insect faction is that insects, by possessing an exoskeleton, have the ability to compartmentalize their olfactory receptor neurons in thousands of sensory hairs on the antenna, thereby giving insects the potential to be able to discriminate closely intertwined, separate odor plumes in ways that humans cannot. Male moths need to be able to optimally receive sex pheromone molecules from females of their own species and discriminate these good pheromone signals from those that contain behavioral inhibitors emitted by females of other species. We propose to perform research to determine how the receptor neurons on moth antennae that are tuned to different sex pheromone components and inhibitors have been placed by evolutionary pressures so that they are co-compartmentalized in the olfactory hairs. By conducting various experiments in which we control the arrival times of different pheromone components and inhibitors, we will be able to determine how much of the fine-grained odor plume resolution ability of the antenna is accomplished by the hairs' arrangement allowing the moth to be able to resolve spatial differences in the arrival of filaments, and how much is accomplished by the neurons in the hairs being able to resolve temporal differences. This research will help us understand how to use pheromones of several species in the same fields at the same time for mating disruption so that the dispensers don't interfere with each other's ability to attract and desensitize the males of each targeted species.

2000-02841 Surface Wax Variation of Peas and Biological Control of the Pea Aphid
Eigenbrode, S.D.
University of Idaho; Department of Plant, Soil and Entomological Sciences; Moscow, ID 83844-2339
Grant 2001-35302-09979; $163,000; 2 Years

This project will examine the potential for genetically altering crop surface waxes to improve biological control of insect pests. Specifically, the research will extend previous work showing that genetically reduced surface wax bloom on peas increases attachment to the plant, and improves mobility and effectiveness of ladybird beetles and lacewing larvae that feed on pea aphids. Consistent with this, the apparent resistance of reduced wax peas in the field to aphids depends on predators; excluding these predators from these peas in the field eliminates the apparent resistance of these peas to aphids. The current project will extend the range of predators studied by determining whether the increased mobility of foliar-foraging predators on reduced-wax peas is synergistic with ground-beetles, which feed on aphids dislodged from the crop canopy. The project will also examine how reduced wax on peas increases attack on the aphids by parasitic wasps. The insect work will include laboratory and field bioassays and behavioral studies. Finally, our pioneering work in this area on peas was conducted with a wax mutation designated 'wel', which has been found to confer a yield disadvantage. As part of this proposal, the effect of the wax mutation 'wsp' on natural enemies of aphids will be assessed in the laboratory and in the field. The principles discovered will be used to guide an effort to incorporate suitable natural enemy-enhancing traits into cultivated pea varieties. Because plant waxes affect beneficial insects on other crops, the research potentially has wider application for improving biological control in agriculture.

2000-02839 The Role of Guarding Behavior in Colony Stinging Responses of Both European and Africanized Honey Bees
Hunt, G.J.; Guzman-Novoa, E.
Purdue University; Department of Entomology; West Lafayette, IN 47907- 1158
Grant 2001-35302-10137; $137,000; 2 Years

Africanized honey bees respond with a much stronger stinging response in defense of their colonies than do European honey bees commonly used in apiculture. Africanized bees are now present near important U.S. queen breeders that are important for the beekeeping and pollination industries, and many beekeepers in these areas now keep Africanized bees. Since honey bee queens mate with a number of males (10-17 drones), these beekeepers have colonies with mixtures of European and Africanized bees. We will determine how European and Africanized bees interact in mixed colonies and how colony defense is organized in these two subspecies. We previously mapped the location of several genes that influence the stinging behavior of Africanized bee colonies. We now seek to make the connection between colony-level and individual-level behaviors. To do this, we need to know how the individual bees of high and low-defensive genotypes interact in mixed-genotype colonies. Preliminary studies indicate that bees converge in likelihood to sting when present in mixed-genotype groups. But interactions between individuals appears to exaggerate differences in guarding behavior. We propose to study individual behaviors in small groups and experimental colonies of both pure and mixed genotypes. We will gather information about the organization of colony defense in both European and Africanized honey bees, and in mixed colonies. There may be a pheromone that modifies likelihood to sting in mixed groups. Knowledge about such a pheromone could lead to methods for chemical modification of stinging behavior.

2000-02978 Molecular Genetics of Hessian Fly Virulence to Wheat

Stuart, J.J.
Purdue University; Department of Entomology; West Lafayette, IN 47907
Grant 2001-35302-09982; $200,000; 3 Years

The Hessian fly is the most important insect pest of wheat. Hessian fly resistance genes in wheat provide an effective method of Hessian fly control and lessen the need for tillage. However, the effectiveness of each resistance gene has a limited duration. Through the action of specific avirulence genes in the Hessian fly genome, Hessian fly populations eventually adapt to overcome resistance, limiting the useful life of each resistance gene to a period of about 8 years. To extend the period of usefulness, we need to discover how Hessian fly avirulence genes function. Toward that goal, this investigation will clone avirulence gene vH13. This gene allows Hessian flies to overcome the resistance conferred by resistance gene H13 in wheat. Chromosome walking will be performed to clone all of the DNA extending between two DNA markers flanking vH13. This DNA will be isolated as a series of overlapping DNA fragments maintained in bacteria. The second objective is to identify vH13 within this series of cloned DNA fragments. This will be accomplished using high-resolution genetic recombination analysis. The structure of the mutations in vH13 that enable it to allow Hessian flies to overcome resistance will be determined by comparing the gene's DNA sequence and its pattern of expression in H13-virulent and H13-avirulent Hessian flies. vH13 will be the first avirulence gene cloned and sequenced from an insect. This investigation will provide knowledge relevant to other avirulence genes in the Hessian fly, and other plant-insect interactions.

2000-02976 Molecular Genetics of Enzymes and Structural Proteins of Aphid Saliva

Reeck, G.R.; Reese, J.C.; Katsar, C.S.
Kansas State University; Department of Biochemistry; Manhattan, KS 66506
Grant 2001-35302-09978; $190,000; 2 Years

The goal of this project is to expand our knowledge of the roles of individual proteins in aphid saliva using tools of molecular biology, which have yet to be applied to this problem. Physiological and biochemical studies have established the importance of salivary secretions in the attack of aphids on plant tissue and have identified several enzyme activities in salivary secretions. There is now a need to begin to individually characterize and study the enzymes and other proteins of aphid saliva and their individual roles in the attack on plant tissue. This has proven intractable at the biochemical level, and we propose to use tools of molecular biology to solve this problem. We will initiate our studies with the pea aphid *(Acyrthosiphon pisum)* because of its relatively large size and the ease with which this insect can be dissected. Our focus will eventually shift to the greenbug *(Schizaphis graminum)* because of its economic importance as a serious pest of U.S. wheat and sorghum crops. Greenbugs and other aphids are major pests of crop plants in this country and abroad. Because salivary secretions are known to be important in the attack on plants by aphids, every salivary enzyme and protein identified by this project is a potential target for genetically engineered resistance of plants to aphid attack. It is our hope that the proposed work and extensions of it will provide new methods, not reliant on chemical insecticides, for control of aphids.

2000-02963 Apoptosis as an Anti-Viral Defense Mechanism in Insects
Clem, R.J.
Kansas State University; Division of Biology; Manhattan, KS 66506
Grant 2001-35302-09972; $116,000; 2 Years

Pest insect populations are naturally limited by a number of factors One such factor is the presence of pathogenic agents such as bacteria, fungi, nematodes and viruses. Although all insects are susceptible to infection by one or more pathogenic agents, they are able to resist most agents quite effectively due to a highly effective innate immune system. There is considerable interest in utilizing insect pathogens as biological control agents to safely curtail insect populations while avoiding the harmful ecological effects of chemical pesticides. However, little is known about the molecular mechanisms utilized by the insect immune system to defeat viruses. An improved fundamental understanding of the insect immune system and how it interacts with viral pathogens will aid in our ability to control pest insects through the development of improved viral biological control agents. All eukaryotic cells possess the ability to commit suicide given the appropriate signal. The genetic pathways that control cell suicide, or apoptosis, are currently an area of intense investigation. It is known that certain insect cells undergo apoptosis upon infection with baculovirus mutants that lack anti-apoptotic genes, and that this premature cell death limits viral replication. It is also known that these baculovirus mutants are have decreased infectivity in insect larvae. Therefore, apoptosis represents a potential anti-viral defense mechanism in insects. This proposal seeks to determine whether apoptosis can, serve as an anti-viral defense mechanism in insects, and to characterize the effects of an apoptotic response on baculovirus pathogenesis in insect larvae.

2000-02970 Role of Baculovirus Expression Factors in Host Range
Passarelli, A.L.
Kansas State University; Division of Biology; Manhattan, KS 66506-04901
Grant 2001-35302-09983; $290,000; 3 Years

Baculoviruses are widespread naturally occurring viruses with large double stranded DNA genomes that infect arthropods in marine and terrestrial ecosystems. They are being developed as biological control agents and thus can benefit ecosystems by reducing the use of chemical pesticides. The host range of each baculovirus species is relatively small, an undesirable property for controlling mixed populations of pest species. The long-term goal of this research is to identify and characterize genetic host range determinants in order to successfully manipulate baculovirus host restrictions and improve their pesticide efficacy. A number of genes (late expression factors or *lefs)* of the baculovirus *Autographa californica* multi-nucleocapsid nucleopolyhedrovirus (Ac*M*NPV) are necessary for late gene transcription and/or DNA replication. Several of these *lefs* have also been associated with host specificity. The specific objectives of this study are to 1) identify *lefs* that play roles as key host range factors, and 2) use site-directed mutagenesis to define the specific function of *lefs* in gene transcription.

2000-02926 Feeding-Induced Resistance by Potato Leafhopper and its Impact on Colorado Potato Beetle
Denno, R.F.; Dively, G.P.
University of Maryland; Department of Entomology; College Park, MD 20742
Grant 2000-35302-9334; $234,000; 3 Years

Colorado potato beetle (CPB) and potato leafhopper (PLH) are both major pests of potatoes. Preliminary evidence suggests that previous feeding by PLH induces physiological changes in potato foliage that negatively affect the performance and survival of CPB which feeds later in the season on the same plants. Consequently, PLH and CPB appear to compete with one another via induced plant resistance. Moreover, because induced plant resistance can delay the development of a later-feeding herbivore, its risk of attack from natural enemies may be enhanced as well. consequently, early-season infestation of potato by PLH, although it can have direct negative effects on potato yield, may have delayed benefits measured in terms of decreased beetle density. Notably, such interactive effects are not currently considered in making management decisions for PLH or other pests in most cropping systems. Using a combination of experiments, the research proposed here addresses two major objectives. First, we thoroughly investigate the PLH-CPB interaction and take advantage of the ideal opportunity it offers to study both the direct and indirect effects of induced plant resistance. Second, we develop the framework for a contemporary economic injury level and set of management decision rules for PLH which reflect the benefits derived from induced resistance to CPB. Our underlying goal with the incorporation of induced plant resistance into management decisions is to reduce the need for insecticide treatments for both pests, which simultaneously lowers the risks of resistance development and enhances the potential mortality from natural enemies.

2000-02832 Genes Related to Pathogenesis in a Fungal Entomopathogen

St. Leger, R.I.
University of Maryland; Department of Entomology; College Park, MD 20742
Grant 2001-35302-10141; $189,000; 3 Years

The new genomic technologies offer unprecedented opportunities for exploiting genes from insect pathogens such as *Metarhizium anisopliae* to engineer insect resistance in plants and to improve biopesticides. We will employ native strains with broad or narrow host ranges, mutants disrupted in key regulators of gene expression and thousands of cloned genes in a microarray survey of gene expression during infection processes, to: 1) identify genes that coordinate the production of infection structures and sundry enzymes/toxins; 2) identify genes involved in determining host range, 3) investigate physiological responses to growth in different insects, and 4) investigate evolution in action by determining if repeated passages through different hosts leads to genetic variants. This project will provide great insight into: 1) the number, nature and networking of genes that regulate and execute infection processes; 2) the intimate relationships between pathogen genes and the host environment, and 3) will identify key targets for precision alterations of pathogen performance. Insect pathogenic fungi represent an untapped reservoir of insect toxins for producing insect resistant plants. We are also proposing to test the biocontrol potential of two proteins that are acutely toxic to caterpillars and beetles. We will: 1) isolate the protein products of these genes from a yeast expression system; 2) examine their pathological effects to determine sites of action, and 3) investigate synergism of toxicity by comparing the effects of the proteins singly and in combination with Bt toxin or an insect diuretic hormone. The impact of this project is expected to extend far beyond *M. anisopliae* in providing a model for-host-pathogen interactions, and provide new and important resources for crop-protection.

2000-01123 Post-translational Processing of Insect Neuropeptide Precursor

Ma, P. W. K.
Mississippi State University; Department of Entomology and Plant Pathology; Mississippi State, MS 39762
Seed Grant 2000-35311-10191; $50,000; 1 Year

The overall project has two objectives: 1) To study the structure and function of insect neuropeptides, and 2) To improve crop quality and productivity and the sustainability of agriculture by developing novel methods for controlling insect pest. The proposed research concentrates on studying the biosynthesis of a neuropeptide, Hez-PBAN (pheromone biosynthesis activating neuropeptide), that is found in the corn earworm, *Helicoverpa zea.* Hez-PBAN is responsible for stimulating the production of sex attractant from many moth species. In addition, the gene that encodes Hez-PBAN also encodes other neuropeptides that function in darkening of larval cuticle, contraction of muscles, and also stimulating physiological dormancy in eggs. By characterizing the peptide fragments that are present in individual neurons, the research will reconstruct the various steps on how a large precursor is being broken down into smaller, physiologically functional molecules. The information gained will be used in the future to develop inhibitors that could interfere with neuropeptide precursor processing in pest insects.

2000-03004 Mechanism of Action of the Insecticidal Toxin Cyt1A: Biophysical and Biochemical Approaches

Butko, P; Russo, P.S.; Pusztai-Carey, M.
University of Southern Mississippi; Department of Chemistry and Biochemistry; Hattiesburg, MS 39406-5043
Grant 2001-35302-10138; $196,000; 3 Years

Toxins from the soil bacterium *Bacillus thuringiensis* (Bt) are a major environmentally safe alternative to chemical insecticides. The object of our study is the toxin Cyt1A, a product of *B. thuringiensis* var. *israelensis*. This toxin is of interest to U.S. agriculture since it is effective against members of the fly family, such as mosquitoes and black flies, which are nuisance pests and carriers of animal diseases. When used in combination with other Bt toxins, Cyt1A synergistically increases efficacy and aids in overcoming insects' resistance to the other toxins. In spite of its commercial use and despite a number of research studies published in scientific literature, Cyt1A's mechanism of action has not been delineated. Taking advantage of a recently solved 3D structure of the related toxin Cyt2A, we will conduct experiments with purified Cyt1A and model cell membrane systems, aimed at elucidating the toxin's mode of action at the molecular level. Using biochemical and biophysical techniques, we will: study the conformation changes in Cyt1A that may be responsible for its interaction with the cell membrane; test by direct methods whether Cyt1A inserts into the membrane or only adsorbs to its surface; and determine the size and morphology of the Cyt1A/lipid complexes. Knowledge of the action mechanism of Cyt1A may bring about novel formulations of Bt-based pesticides with increased insecticidal efficacy. It also may provide hints for enhancing its specificity for target insects and improving the safety of non-target organisms, including humans.

2000-01195 Centrifuge for Research on Herbivore-Elicited Pigment and Protein Changes of Plants

Ni, X.; Quisenberry, S.S.
Montana State University; Department of Entomology; Bozeman, MT 59717
Equipment Grant 2001-35311-10193; $16,695; 1 Year

Requested funds will be used to purchase a refrigerated high-speed centrifuge with three rotors. This instrument will significantly upgrade our research capabilities by allowing us to conduct density gradient centrifugation, and perform routine highspeed centrifugation protocols under refrigerated condition. Our current research isolates wheat chloroplasts and monitors aphid-elicited chlorosis. The centrifuge will allow us to isolate large quantity of wheat chloroplasts using Percoll density gradient centrifugation. The instrument will also allow us to conduct refrigerated high-speed centrifugation that is critical for processing both insect and plant enzyme samples. The centrifuge will not only play a key role in successful completion of the current research projects, but also upgrade the infrastructure of basic research in our department and increase the competitiveness of our department to effectively pursue extramural funding in the future.

2000-01146 Gas Chromatograph/Mass Spectrometer for Tritrophic Insect Chemical Ecology Research

Weaver, I.K.; Morrill, W.L.; Johnson, G.D.
Montana State University; Department of Entomology; Bozeman, MT 59717
Equipment Grant 2001-35311-10121; $49,797; 1 Year

Requested funds will be used to purchase a capillary gas chromatograph/mass spectrometer. This instrument will strongly enhance our research efficiency by allowing us to conduct in-house routine chemical analyses of collected volatile samples from biological preparations, and from synthetic blends on substrates. Our research deals with cereal pests that induce chemical changes in host plants as they are feeding, the responses of natural enemies to these volatile signals and with chemical communication within the insect species involved. The gas chromatograph/mass spectrometer will not only be vital to the timely completion of our ongoing research, but will also upgrade the infrastructure of basic research in our department, and thus enhance our competitiveness in obtaining extramural funding in future competitive proposals.

2000-02931 Thrips and Tomato Spotted Wilt Virus: Interactions of Temperature, Host & Virus

Kennedy, G.G.
North Carolina State University; Department of Entomology; Raleigh, NC 27695-7630

Grant 2001-35302-09885; $176,000; 2 Years

Tomato spotted wilt virus (TSWV) has recently emerged as one of the most devastating insect transmitted plant viruses affecting food, fiber, and ornamental crops world-wide. TSWV is transmitted by at least 8 species of thrips. In the USA, 2 species, the tobacco thrips and the western flower thrips, are the most important vectors of TSWV. Unlike most insect-transmitted plant viruses, TSWV actively infects its thrips vectors and its plant hosts. Although a great deal is known about the molecular biology of TSWV, our understanding of the factors influencing the spread of TSWV is very limited. This limited understanding is impeding our ability to predict and to manage outbreaks of TSWV. This research will examine how temperature interacts with plant species and different virus isolates to influence the number of infectious tobacco thrips and western flower thrip produced on TSWV-infected plants. Additionally, the research will examine thrips populations from a number of different geographic locations to determine if there are differences in the way temperature, TSWV isolate and host plant affect their population growth and transmission of TSWV. This research will help to link our knowledge of the molecular and cellular level effects of TSWV on its thrips vectors to the patterns of variation that exist in the timing and intensity of TSWV outbreaks in agricultural crops. The results should help us to explain shifts in the relative importance of particular thrips species as vectors of TSWV and in the importance of particular strains of the tomato spotted wilt virus.

2000-02827 Colony and Population Genetic Structure of the Eastern Subterranean Termite

Vargo, E.L.
North Carolina State University; Department of Entomology; Raleigh, NC 27695

Grant 2000-35302-9377; $159,000; 2 Years

Subterranean termites of the genus *Reticulitermes* are destructive pests throughout most of the U.S., where they damage buildings including many structures important in agriculture. In addition, in many areas of the world, various subterranean termite species are serious pests of a number of crops and timber trees. Information on subterranean termite social structure and colony spatial organization is critical to the development and implementation of more effective methods of control, especially with the growing interest in baiting technologies. However, the cryptic nesting and foraging habits of these species have made it difficult to obtain such information. The proposed work will combine data from two classes of molecular genetic markers to conduct detailed studies of colony and population genetic structure, as well as the size of foraging areas occupied by the eastern subterranean termite *R. flavipes*. The main objectives of this work are: 1) to determine the predominant type of breeding system (the numbers and genetic relatedness of reproductives present in colonies); 2) to determine whether the major features of the breeding system are consistent within habitats and vary between habitats; 3) to determine patterns of gene flow within and among areas; and 4) to determine the relationship between colony social organization and size of foraging area. Through a better understanding of the social organization, breeding system and feeding ranges of subterranean termites, the proposed work will make a significant contribution toward the development of new and improved methods of subterranean termite control.

2000-02923 Strategies for Identifying Targets to Insect Ecdysteroid Receptors
Henrich, V.C.
University of North Carolina, Greensboro; Department of Biology; Greensboro, NC 27402
Grant 2000-35302-9327; $201,000; 2 Years

These studies will focus on the development of rational approaches for the identification of novel insecticides. The target of these studies is a protein receptor that normally processes steroid hormone signals produced by the insect. These signals regulate the progression of insects through larval feeding stages to adults. It is already known that this receptor, which exists in virtually all insects, can be blocked or activated abnormally by specific chemical agents, so that insect development is disrupted, resulting in death. These experiments utilize both cell cultures and transgenic insects to develop assays that in turn, will be used to test known and novel agents for their ability to activate or impair receptor response. Further, the receptors of different insects will be compared for their responsiveness to specific compounds in an effort to identify potential species-specific pesticides. Finally, by producing altered forms of these receptors and testing their potential for response, subtle structural differences between insect steroid hormone receptors will be identified that are associated with differences in responsiveness to candidate insecticides.

2000-03024 Tree Defenses, Community Interactions, and the Population Dynamics of *Dendroctonus frontalis*
Ayres, M.P.
Dartmouth College; Department of Biological Sciences; Hanover, NH 03755
Grant 2001-35302-09921; $225,000; 3 Years

Understanding why some species regularly attain very high abundance is a central problem in theoretical population ecology and applied entomology. We will test for the contributions of two biological processes to the outbreak population dynamics of the southern pine beetle: (1) positive density--dependence from effects of beetle attack rate on tree defense systems; and (2) negative density-dependence from community interactions involving mutualistic fungi that aid in beetle nutrition, a bluestain fungus that competes with the mutualistic fungi, and mites that travel on the beetles and propagate the bluestain fungus. The first process would increase the peaks in population abundance while the second would limit the duration of outbreaks. We will characterize variation in tree defenses within and among pine species and parameterize a model that predicts future outbreaks as a function of tree defenses and present abundance. The model will be validated through field studies and then extended to predict outbreak probabilities under different scenarios of forest composition and tree physiology. This will aid forest managers by permitting population predictions from measurements that are cheap and easy. We will test the role of fungal interactions with a combination of experiments and field held surveys that will tell us if this feedback contributes to cycles of beetle outbreaks. Finally, we will integrate our new data with existing knowledge of density-dependence from intraspecific competition and specialist predators to develop synthetic models and test whether combined effects from different sources of feedback produce population dynamics that differ from those produced by individual components.

2000-02822 A Potent Feeding Stimulant for *Manduca sexta:* Identification, Occurrence and Function
Renwick, J.A.A.
Boyce Thompson Institute for Plant Research; Ithaca, NY 14853
Grant 2001-35302-09884; $161,000; 2 Years

Many insect pests of specific crops are specialists on a particular plant family, and they utilize characteristic chemicals in these plants to recognize their hosts. But the chemistry responsible for such close plant-insect associations is largely unknown. Information about the chemical dependence of pest insects is needed to guide the design of resistant plants and to provide leads for new, environmentally acceptable pest management strategies. The Colorado potato beetle, *Leptinotarsa decemlineata,* and the tobacco hornworm, *Manduca sexta,* have long served as prime examples of insects that are stimulated to feed by juices or extracts from their host plants. But the specific chemicals that trigger the behavioral response have remained elusive. Recent work in the PI's laboratory has led to the discovery of a potent feeding stimulant from potato for larvae of M. *sexta,* and these larvae become dependent on this compound for further feeding. This research is designed to identify the compound involved and to determine whether the same or different compounds are responsible for host recognition by the potato beetle. Behavioral assays will be used to monitor isolation of the active compounds, and a combination of spectroscopic techniques will be used for chemical identification. Methods will be developed for detection of these key chemicals in different varieties and species of plant. Monitoring levels of these chemicals is expected to facilitate the selection of crop lines that lack the stimulant, and plants with high levels of stimulant might serve as a trap crop for natural pest control.

2000-02955 Physical and Chemical Determinants of Varroa Mite Invasion of Honey Bee Brood Cells

Calderone, N.W.
Cornell University; Department of Entomology; Ithaca, NY 14853-0901
Grant 2001-35302-09889; $126,000; 2 Years

The honey bee, *Apis mellifera,* provides pollination for over 90 commercial crops in the U.S. The value of increased production attributable to honey bees is estimated to be $14.5 billion/year. The parasitic bee mite *Varroa jacobsoni* was detected in the US in 1987 and poses a grave threat to the US beekeeping industry and to sectors of the agricultural community that produce fruits and vegetables requiring honey bees for pollination. Untreated, *Varroa* is usually fatal to honey bee colonies within 2 years of infection. Apistan is registered for *Varroa* control, but resistance has become widespread in the past 2-3 years. CheckMite+ received an EPA Section 18 registration with 'non-food use' status; however, several factors, including the recent finding of *Varroa* resistance to Coumaphos, make it an unattractive opinion. A comprehensive understanding of the host-parasite relationship is necessary to enable the development of additional control strategies. During the past 2.5 years, my lab has investigated the mite's host location process, a process that is critical to the mite's ability to successfully reproduce. Our understanding of the mechanisms underlying that process has now reached a mature state. Recently, we identified physical and chemical stimuli that affect the host location process. We propose to examine these stimuli in greater detail. Results from these studies will play a key role in the development of a mite trap and in efforts to disrupt the mite's host location process.

2000-02829 Targeted Ablation of Developmentally-important Peptidergic Neurons in *Drosophila*

Ewer, J.
Cornell University; Department of Entomology; Ithaca, NY 14853
Grant 2001-35302-10140; $175,000; 2 Years

Insect growth from larva to adult occurs through multiple stages. At the end of each stage insects molt to produce a new cuticle for the next stage. During this process, the new cuticle develops beneath the old one, while much of the old cuticle is resorbed. The final, vital, step of this developmental process is ecdysis, the shedding of the remaining old cuticle. Research conducted in the hornworm, *Manduca sexta,* has revealed that the neuropeptide Crustacean CardioActive Peptide (CCAP) is involved in turning on ecdysis behavior. The goal of this study is to use the fruit fly, *Drosophila melanogaster,* to demonstrate that CCAP is the neuropeptide that activates ecdysis behavior in the intact animal. The advantage of *Drosophila is* that the genetic and molecular tools available in this organism make it possible to investigate in detail the role of CCAP *in vivo.* To do so, we will examine the ecdysis behavior of animals in which the CCAP neurons have been selectively killed using transgenic animals. The proposed research will contribute to our understanding of the role of CCAP in the control of this essential insect behavior. It may also lead to the development of powerful agents for insect control. Indeed, successful ecdysis is vital to the survival and growth of insects. Thus, agents that interfere with ecdysis would be lethal. They would also kill the insect at an early stage, thus preventing the animal from reaching the larger, more crop-damaging stages.

2000-03008 Genetic Diversity and Disease Within Honey Bee Colonies

Tarpy, D.R.
Cornell University; Department of Neurobiology and Behavior; Ithaca, NY 14853-2702
Postdoctoral Fellowship; Grant 2001-35302-09905; $89,980; 2 Years

Honey bee colonies are subject to a wide range of parasites and pathogens. The honey bee mating system, where queen bees may mate with as many as 20 males, has been suggested to reduce these high disease loads within colonies by increasing the genetic diversity among colony members. As mating number increases for the queen, the variation in disease-resistant genes increases among the worker population, which may cause parasites and pathogens to repeatedly adapt to the different hosts' genotypes. The consequence may be a reduced transmission rate of disease among colony members, increasing the chances of colony survival. This hypothesis makes several predictions that can be tested empirically. First, the variance in worker survival should be smaller among genetically-diverse colonies compared to genetically-uniform colonies. Second, the transmission rate of disease should decrease as a function of increasing genetic diversity. The objective of this proposal is to test these predictions in two separate experiments. The first experiment compares non-commercial colonies that vary in their genetic complexity for differences in worker populations and prevalence of disease. The second experiment tests the effects of both brood and worker diversity on the rate of pathogen transmission, simultaneously investigating multiple mechanisms of disease resistance. Together, the two studies will demonstrate if multiple mating influences colony survival by the mechanism proposed in the hypothesis. If increased genetic diversity benefits colony survival, it may be advantageous for the apicultural industry to maximize queen mating numbers to alleviate the high levels of disease that plague commercial colonies.

2000-02942 Structure/Function of Pheromone Biosynthetic Enzymes of Lepidopteran Pests

Knipple, D.C.
Cornell University; New York State Agricultural Experiment Station; Department of Entomology; Geneva, NY 14456
Grant 2001-35302-09926; $202,000; 3 Years

The long-term goal of this project is to develop basic knowledge and materials necessary to increase the use of highly specific, non-toxic sex pheromones as alternatives to conventional toxic insecticides for the control of moth species that are major agricultural pests. Despite their advantages, pheromones have had limited commercial success due to their high synthesis costs. The development of cheaper, biologically-based syntheses of pheromones is now feasible with genetic technology, but will require the control of key biosynthetic enzymes. Toward this goal, this project will isolate and functionally express moth DNA sequences encoding two important categories of pheromone biosynthetic enzymes: acyl-CoA desaturases and b-oxidation enzymes. The many unique functional properties among these two categories of enzymes in moth sex pheromone glands account for much of the diversity of chemical structures used as sex pheromones in this group of insects. The biological materials that will provide sources of DNA for these investigations will come from agriculturally important pests such as the corn ear worm, the tobacco bud worm, the pink boll worm, the cabbage looper, and the codling moth. The specific experimental objectives are to functionally express these enzymes in yeast cells, to characterize their functional properties and the structural elements conferring them, and to construct enzymes with improved properties. Accomplishment of these goals will contribute to basic understanding of the moth pheromone biosynthesis system and will develop potential tools to produce less expensive pheromone products, which can be used to improve agricultural productivity and profitability through environmentally benign means.

2000-03016 Microparticle Dispensers for the Controlled Release of Insect Pheromones
Stipanovic, A.J., Webster, F.X.
State University of New York; College of Environmental Science and Forestry; Syracruse , NY 13210
Grant 2001-35302-10082; $112,000; 2 Years

Over the past several decades, it has been firmly established that the primary mechanism of communication among insects involves chemical compounds called pheromones. Although these molecules are generally released by insects in very low concentrations, they play a vital role in mating and other functions. Once a pheromone is isolated and its chemical structure elucidated, synthetic analogs can be used to control the mating behavior of specific insects. To exploit pheromones in pest management, it is necessary to incorporate these synthetic analogs into a controlled release device from which they can evaporate slowly at a rate similar to the natural emission of female insects. If sufficient pheromone sources are distributed in the environment, male insects become confused and cannot find mates. In this study, we will develop micron-sized control release particles that can be sprinkled in orchards and forests to control several agricultural pests including the codling and gypsy moths. Those micro-dispensers will have several advantages over existing controlled release devices: they will be relatively inexpensive, easy to implement in the field and they will be biodegradable. This study will also investigate, from a fundamental molecular perspective, how pheromones diffuse through solid matrices and how they evaporate into the atmosphere. It is anticipated that a model can be generated using “Neural Networks" that predicts the release rate of pheromones from relatively simple physical and chemical measurements.

2000-02843 Longevity and Stress Tolerance of Infective Juvenile Entomopathogenic Nematodes
Grewal, P.S.
Ohio State University; Department of Entomology; Wooster, OH 44691
Grant 2000-35302-9336; $156,000; 2 Years

Entomopathogenic nematodes (Heterorhabditidae and Steinernematidae) are already used in biological control of insect pests of citrus, turfgrass, strawberries, cranberries, mushrooms, and greenhouses. Further expansion of their use is hampered by limited shelf-life and poor field persistence due to short longevity and intolerance to environmental stresses. Research is proposed to elucidate mechanisms of aging and stress tolerance in infective juveniles of the entomopathogenic nematode, *Heterorhabditis bacteriophora*, to develop novel methods providing improved storage stability and field persistence. Recombinant inbred lines with extremes of infective juvenile longevity and stress tolerance will be isolated from a newly established F-2 hybrid between 'long-lived' and 'short-lived' strains of *H. bacteriophora.* The inbred lines will be used to test several hypotheses referred to as the 'theories of aging'. Specific objectives are: (i) Determine correlations between longevity and tolerance to environmental stresses including heat, freezing, hypoxia, desiccation, and UV in recombinant inbred lines, (ii) determine physiological and molecular differences in the inbred lines with extremes of longevity and stress tolerance, (iii) determine differences in the infectivity and field persistence of the inbred lines with extremes of longevity and stress tolerance. This study will identify long-lived inbred lines of *H. bacteriophora* that possess high tolerance to major environmental stresses. Likelihood of ultimate success is greatly enhanced because a closely related and extensively studied nematode *Caenorhabditis elegans* serves as a model for understanding the genomic architecture of entomopathogenic nematodes.

2000-02914 Insect Adaptation to Plant Defense: Decryption of Gut Protease Deployment, and Development of Synthetic Resistance
Zhu-Salzman, K.
Texas A&M University; Department of Entomology; College Station, TX 77843
Grant 2001-35302-10136; $198,000; 3 Years

Significant agricultural loss is caused by insect damage. One of the best methods for managing insect pests is to plant cultivars that express insect resistance genes. Transformation techniques allow introduction of new resistance genes into plants, for example *Bacillus thuringiensis* (Bt) toxin-mediated insect resistance in crops has become a successful, widely used control measure. However, insects are occasionally observed to overcome this resistance, necessitating a continuing search for other protein toxins. Various anti-insect genes have been isolated, including plant protease inhibitors. Although protease inhibitors have been shown to powerfully inhibit digestion in some insects, successful control via expression of these inhibitors in transgenic plants has been very limited. Scientists have gradually realized that our lack of understanding of insect physiological and biochemical systems that are vulnerable to resistance genes, and of the molecular basis of co-evolution between insects and plants, are a barrier to the success of biotechnology-based approaches for insect pest management. If genetic engineering of crop plants for insect resistance is to become a more widely applicable tool for insect pest management in the future, we must understand how insects cope with plant defense proteins. Accordingly, the goal of this project is to investigate insect counter defense mechanisms, using a soybean protease inhibitor and the cowpea bruchid as model systems. The project will design defense molecules that can delay insects' development of resistance. Information gained about plant-insect interactions can facilitate development of genetic engineering strategies to improve plant resistance, offering great potential benefit to U.S. agriculture.

2000-02873 Translocation of Herbivore Elicitors and Induction of Plant Volatile Emissions

Paré; P.W.
Texas Tech University; Department of Chemistry and Biochemistry; Lubbock, TX 79409
Grant 2000-35302-9378; $199,000; 3 Years

Plants normally have little odor associated with their leaves, however when a plant is damaged by a leaf eating insect, the odor given off from the foliage becomes much stronger. These odors attract natural enemies of the feeding insect; in turn, these beneficial insects, by stinging, can terminate the reproductive cycle for the herbivore. Key to the emissions of these volatile plant attractants are activating substances present in the oral secretion of the herbivore. The first such chemical elicitor that triggers the production of plant volatiles has recently been identified from the beet armyworm. We propose to radiochemically label this elicitor of plant volatile emissions and track the movement of this single molecule in the plant. This tracking of chemical components from the caterpillar to the plant will be essential for establishing how insect components signal the occurrence of herbivore damage in plants. The pattern of volatile emissions in on undamaged portion of the plant will also be monitored. Herbivore elicitors that trigger a plant's natural defenses against crop pests is a promising strategy to reduce chemical insecticide usage. For sustained plant protection, activation of indirect defenses such as the attraction of natural enemies, along with defenses that are aimed directly at the herbivore, will play an important role in crop protection.

2000-02916 Cellular Modulation of Complex Behaviors; Ecdysis Behavior in Insects

Fuse, M.M.
University of Washington; Department of Zoology; Seattle, WA 98195
Postdoctoral Fellowship; Grant 2001-35302-09985; $90,000; 2 Years

Insects are major agricultural pests which cause substantial losses to agriculture and forestry industries. Identifying roles of insect-specific peptides involved in the regulation of insect development should be beneficial for the design and implementation of novel pest management strategies. Ecdysis, or the shedding of the old cuticle, occurs at the end of each molt in insects, and is essential for growth and development. This project will characterize the neural control of ecdysis using the moth, *Manduca sexta.* Specifically, the neurons involved in controlling the timing of ecdysis, and novel peptides involved in ecdysis, will be identified. A model for the interactions between various neuropeptides identified in ecdysis has been proposed for coordinating pre-ecdysis and ecdysis behaviors. Ligature experiments in both moths and flies have suggested that some aspects of timing of these events depend on descending inhibitory inputs from the head. This project will look at the cellular mechanisms of modulation of ecdysis behaviors by (i) identifying the inhibitory neurons which coordinate tuning of ecdysis, using electrophysiology techniques, and (ii) characterizing novel peptides involved in ecdysis using histochemistry and bioassays. The ability of modified peptide analogues to cross the blood-brain barrier or cuticle will also be tested, to provide a means of manipulating ecdysis events *in vivo* in future experiments. This work provides a model for studying modulation of neural networks in general, but may also be the basis for the design and implementation of novel pest management strategies.

2000-03009 Regulation of Caste Fate in the Polyembryonic Wasp *Copidosoma floridanum*

Corley, L.S.
University of Wisconsin; Department of Entomology; Madison WI, 53706
Postdoctoral Fellowship; Grant 2000-35302-9401; $90,000; 2 Years

The goal of this proposal is to contribute new knowledge to a fundamental aspect of insect biology--caste specialization. Caste specialization is one of the most important life history strategies to have arisen within the insects. In many social insect species such as ants, bees, and wasps, individual members of a colony are morphologically and functionally distinct. This differentiation results in the production of castes; for example, in bees, sterile workers (drones) assist reproductives (queens) in defense and offspring care. The differences in morphology and function between castes is known to be influenced by interactions between environmental, genetic, and endocrine factors. How these interactions work to produce different castes is not well understood. I have proposed to characterize the developmental and genetic processes underlying caste formation in the parasitoid wasp *Copidosoma floridanum*. *C. floridanum* lays its eggs into the egg stage of its host, the cabbage looper *Trichoplusia ni*. In this species, two distinct larval castes arise from a single egg by a clonal (polyembryonic) developmental mechanism. The results of this project will not only contribute to our understanding of insect caste specialization but will also aid in our knowledge of parasitoid development and life history. Parasitoid wasps are important biological control agents and the proposed work will contribute to our understanding of parasitoid development and its impact on their lifestyles.

2000-02982 Ecological Consequences of Multiple Global Change Factors: CO_2, O_3 and Tree-Insect Interactions

Lindroth, R.L.
University of Wisconsin Madison; Department of Entomology; Madison, WI 53706
Grant 2001-35302-09984; $102,000; 2 Years

Increasing concentrations of atmospheric carbon dioxide (CO_2) and ozone (O_3) are anticipated to exert significant impacts on forest ecosystems over the next 50-100 years. However, the consequences of pollutant exposure for plant-insect interactions, including effects on both insect performance and feedback effects on tree productivity, are poorly understood. Critically needed are ecologically realistic studies of the effects of CO_2 and O_3 on multiple tree and insect species. Such are now possible with Free Air CO_2 Enrichment (FACE) technology. A recently constructed FACE facility in northern Wisconsin is currently the largest FACE system in the world, and the only one addressing the individual and combined effects of CO_2 and O_3 on forests. The purpose of this research is to assess the independent and interactive effects of CO_2 and O_3 on the chemical quality of tree leaves, and the consequences of such changes for rates of defoliation, insect performance, and insect tolerance to insecticides. Experimental species include paper birch *(Betula papyrifera)*, quaking aspen *(Populus tremuloides)*, forest tent caterpillar *(Malacosoma disstria)*, white-marked tussock moth *(Orgyia leucostigma)*, and gypsy moth *(Lymantria dispar)*. This research will improve our understanding of key atmospheric and biological stress factors influencing the health and productivity of forest ecosystems over the 21st century.